

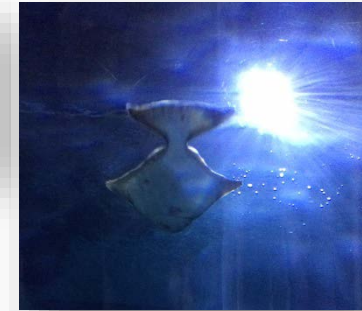


IPHC 5-year Research Program

**RAB Meeting
November 16, 2016**

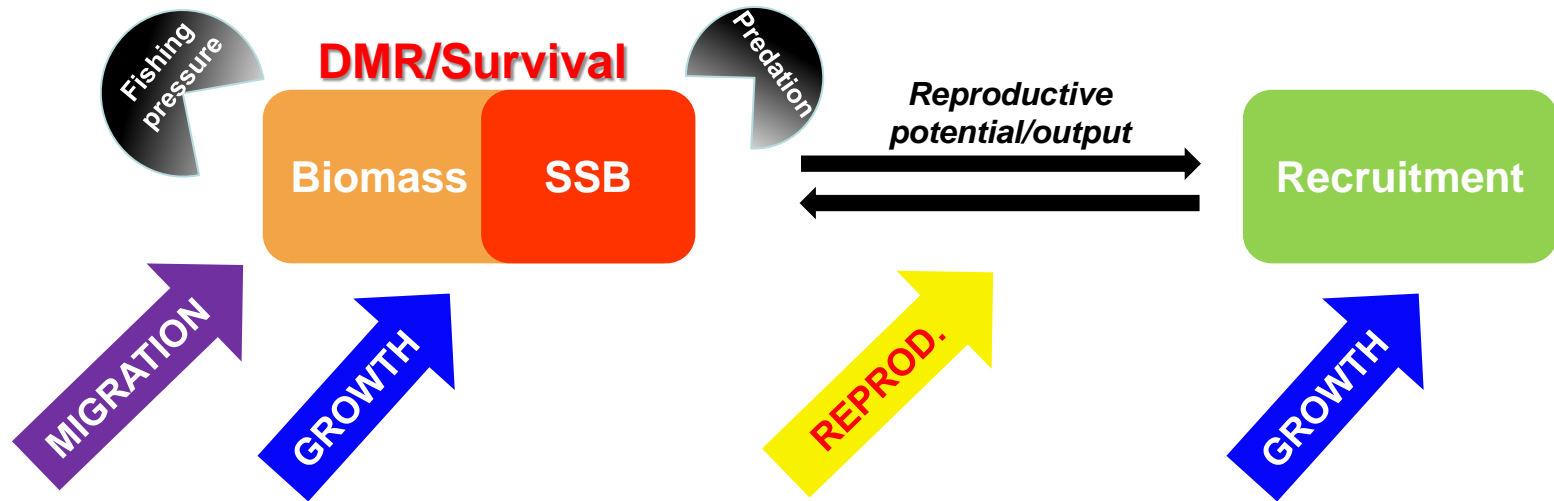
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Program Manager*

Planned research activities at IPHC



Primary objectives

- Identify and address *critical knowledge gaps* in the biology of the Pacific halibut
- Understand the influence of *environmental conditions* on halibut biology
- Apply resulting knowledge to reduce *uncertainty* in current stock assessment models



Planned research activities at IPHC



1. Reproduction

- SEX RATIO OF CATCH
- IMPROVED MATURATION ESTIMATES OF SPAWNING BIOMASS

2. Growth

- CHANGES IN SIZE AT AGE/BIOMASS
- TOOLS TO ASSESS FISH CONDITION

3. DMRs and post-release survival assessment

- BYCATCH SURVIVAL ESTIMATES

4. Migration

- LARVAL DISPERSAL
- ADULT FEEDING AND REPRODUCTIVE MIGRATION

5. Genetics and genomics

- GENETIC STRUCTURE OF THE POPULATION
- GENOMIC TOOLS (e.g. GENOME)



1. Reproduction: proposed studies

There are important knowledge gaps on the reproductive biology of the species

- SEX RATIO OF CATCH
- IMPROVED MATURATION ESTIMATES OF SPAWNING BIOMASS

What is needed?

- Knowledge on reproductive development, maturation, fecundity, sex determination mechanisms (sex identification), environmental and hormonal control of reproduction.
- Scientific-based criteria to identify reproductive status and potential.
- Updated estimates of age and size at maturation.
- Information on skipped spawning.

New proposed studies:

- *Full characterization of the annual reproductive cycle*
- *Identification of sex determination mechanism(s) and influencing factors*



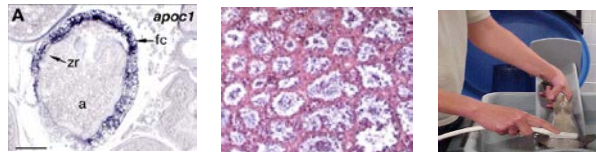
1. Reproduction: proposed studies

- ***Full characterization of the annual reproductive cycle***

Objective: Understand temporal changes in reproductive development throughout an entire annual reproductive cycle in male and female Pacific halibut

- Histological assessment of gonadal development and maturation.
- Endocrine profiling (hormone levels in the blood) of the reproductive cycle.
- Gene expression (transcriptome) profiling of the reproductive axis.
- Gonadosomatic index (GSI) measures throughout the reproductive cycle.
- Ultrasound monitoring of gonadal development and maturation.

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Deliverables:

- Comprehensive reproductive monitoring of the adult population in order to improve our estimate of actual spawning biomass
- Accurate staging of reproductive status
- Updated maturity-at-age estimates.
- Estimates of skipped-spawning



1. Reproduction: proposed studies

- **Identification of sex determination mechanisms**

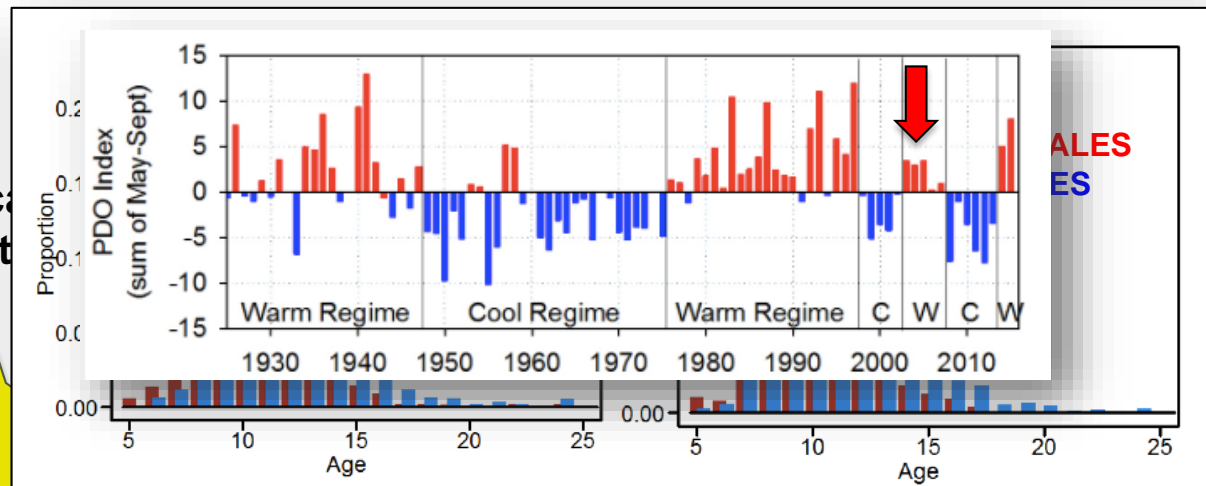
Objective: Understand how sex is established in Pacific halibut

- Identification of sex markers (SNPs)
- Identification of the sex determining mechanism(s) and its onset during early development.
- Identification of environmental influences (e.g. temperature) on sex determination.
- Evaluate possible consequences on sex ratios at the population level.

Deliverables:

- Genetic
- Identific
- Informat

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FY2017



2. Growth: proposed studies

Little is known regarding what factors influence growth in this species

- CHANGES IN SIZE AT AGE/BIOMASS
- TOOLS TO ASSESS FISH CONDITION

What is needed?

- Knowledge on growth patterns and environmental influences.
- Improved understanding in the possible role of growth alterations in the observed decrease in size at age.

New proposed studies:

- *Extensive catalogue of physiological markers to monitor growth*
- *Evaluation of growth patterns and effects of environmental influences*



2. Growth: proposed studies

- **Extensive catalogue of molecular markers for growth**

Objective: Identify and validate molecular growth-related markers for growth studies.

- Identification of expressed sequences from skeletal muscle (white and red) and liver.
- Develop molecular assays to quantify gene expression of growth markers in relevant tissues.



RNA sequencing **Ongoing**

3. De novo Transcriptome Assembly Stats

Sample ID	Total trinity 'genes'	Total trinity transcripts	Percent GC	Contig N50	Median contig length	Average contig	Total assembled bases	
R116-pool1	37,161	39,638	47.76	1,198	385	721.49	28,598,382	WHITE MUSCLE
R116-pool2	38,143	40,814	46.02	1,096	398	691.85	28,237,340	LIVER
R116-pool5	70,693	86,561	47.17	2,104	495	1,051.87	91,050,930	RED MUSCLE

5.1 Mapping statistics

Sample ID	Danio rerio	uniprot	est others	total	unmapped	Danio%	uniprot%	est others%	unmapped%	
R116-pool1	13,873	2,661	23,066	39,638	38	35.00%	6.71%	58.19%	0.10%	WHITE MUSCLE
R116-pool2	13,233	2,547	24,998	40,814	36	32.42%	6.24%	61.25%	0.09%	LIVER
R116-pool5	25,341	5,579	55,466	86,561	175	29.28%	6.45%	64.08%	0.20%	RED MUSCLE

Deliverables:

- Establishment of a growth-related gene sequence dataset
- Molecular assays to monitor growth patterns



2. Growth: proposed studies

- **Evaluation of growth patterns and effects of environmental influences**

Objective: Identify molecular, biochemical and isotopic profiles characteristic of specific growth patterns and evaluate potential effects of environmental influences.

- Evaluation of different growth trajectories in the wild.

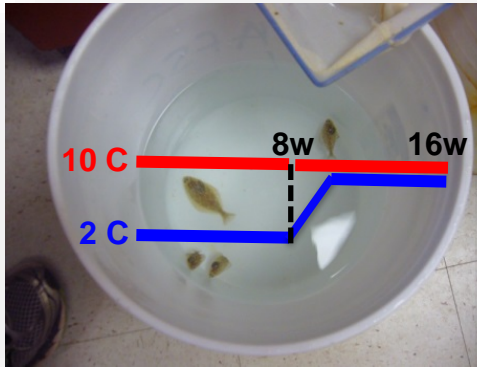
In BS NMFS trawl survey in 2016:

- - 75 fish <40 cm length
- - 75 fish 40-60 cm length
- - 75 fish 60-80 cm length



Characterization of molecular and biochemical growth markers in liver and muscle samples from age-matched individuals

- Establishment of different growth trajectories in juvenile fish in captivity to identify molecular and biochemical signatures of growth patterns.



...s (ration, density, thermal- or fasting-induced

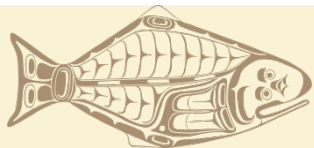
Normal rate

High rate



- Isotopic tissue turnover to trace dietary and/or habitat shifts

^{13}C , ^{15}N



2. Growth: proposed studies

- Investigate the effects of **environmental factors** on growth performance.
 - *Effects of **temperature, salinity, dissolved oxygen** and **water pH** on growth.*
 - *Identify the optimal environmental conditions for growth.*
- Understand the basis of the **sexual dimorphic growth** in the Pacific halibut.

Deliverables:

- Identification and validation of growth markers for field studies
- Characterization of molecular and biochemical growth signatures
- Environmental effects on somatic growth
- Improved biological inputs on biomass estimates



3. DMRs and survival: proposed studies

Little is known regarding what factors influence bycatch survival and how

- BYCATCH SURVIVAL ESTIMATES

What is needed?

- To introduce quantitative measurable factors that are linked to fish handling practices and to fish physiological condition and ultimately to survival in order to improve current DMR estimations

New proposed studies:

- *Evaluation of the effects of fish handling practices on injury levels and the physiological condition of captured Pacific halibut*
- *Investigate the relationship between physiological condition post-capture and survival as assessed by the use of accelerometer tags.*
- *Improving estimates of survival of Pacific halibut caught in the trawl fishery*



3. DMRs and survival: proposed studies

- ***Evaluation of the effects of fish handling practices on injury levels and the physiological condition of captured Pacific halibut***

Objective: Understand relationship between handling practices and physiological condition of captured Pacific halibut in the longline fishery

- **Assess injuries associated with release techniques (gangion cut, careful shake, hook straightening).**
- **Determine the physiological condition of all captured fish with associated injury levels after different deck exposure times: condition factor index (Kn), energy (fat) levels, morphometric analyses.**
- **Measure the levels of stress and physiological disturbance indicators in the blood of all captured fish (cortisol, lactate, glucose, potassium, hematocrit).**

Deliverables:

- **Injury profile for different release techniques in the longline fishery**
- **Physiological assessment of fish handling practices: fish condition index post-capture**



3. DMRs and survival: proposed studies

- *Investigate the relationship between physiological condition post-capture and survival as assessed by tagging.*

Objective: Measure survival post-release in Pacific halibut and link this with the physiological condition and capture-related events

- Tag fish that have been exposed to different handling practices in the longline fishery with accelerometer tags in addition to conventional tags (wire).
- Assess survival of fish according to size and physiological condition.

Deliverables:

- Information on post-release survival in relation to handling practices and physiological condition.
- Information on post-release survival in relation to size.
- Estimating DMRs by EM.



3. DMRs and survival: proposed studies

- *Improving estimates of survival of Pacific halibut in the trawl fishery*

Objective: Assess the survival of discarded Pacific halibut in the non-directed trawl fishery



discarded Pacific halibut in the non-directed trawl fishery to estimate discard mortality rates

- Continue and expand collaborative work with the industry
- Apply methods developed in Amendment 80 fleet to plan
- Determine survival of captured halibut.
- Relate physiological condition of discarded halibut

discarded halibut

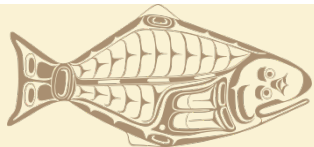
captured halibut.

aging.

discarded halibut

Deliverables:

- Improved knowledge of survival of discarded halibut and, consequently, improved estimates of discard mortality rates in the trawl fishery.



4. Migration: proposed studies

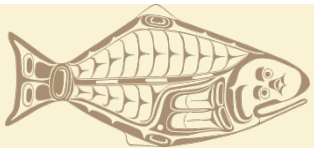
- LARVAL DISPERSAL
- ADULT FEEDING AND REPRODUCTIVE MIGRATION

What is needed?

- Improve our understanding on larval, juvenile and reproductive migration.
- Incorporate additional sources of biological information on migratory.

New proposed studies:

- *Towards a more integrative view on migration*
- *Larval migration and connectivity*
- *Swimming and migratory performance*



4. Migration: proposed studies

- *Towards a more integrative view on migration*

Objective: Combine current tagging efforts with genetic and otolith and tissue composition analyses.

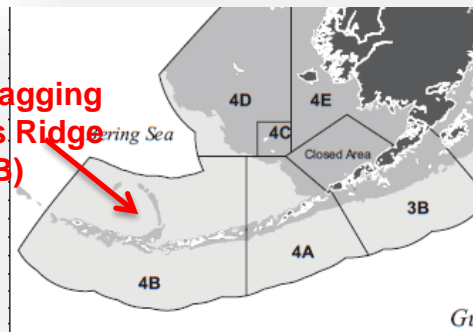
- Genetic analyses of tagged fish to shed light on migration patterns and geographic origin.
- Otolith microchemical and stable isotope analyses and tissue stable isotope analyses.
- Reproductive monitoring of PAT-tagged adult females: blood endocrine reproductive parameters, ovarian tissue biopsies and ultrasound for ovarian staging.

Deliverables:

- Genetic and elemental and isotopic information on migratory adult fish
- Improved knowledge on reproductive migrations and identification of spawning areas



- PAT tagging Bower's Ridge (4B)



- Tail pattern recognition

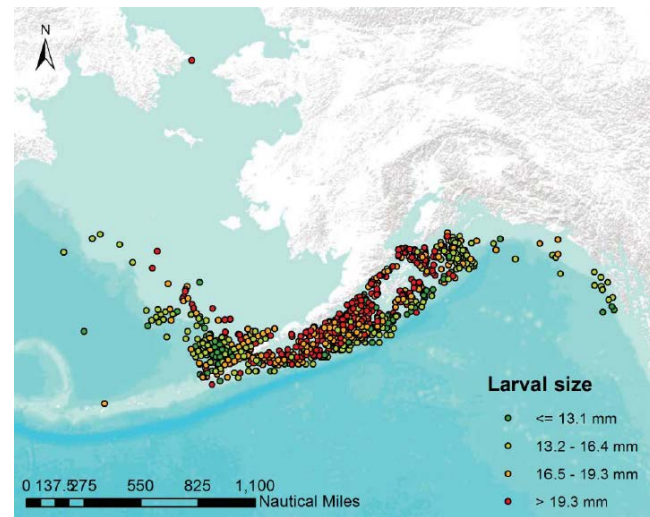
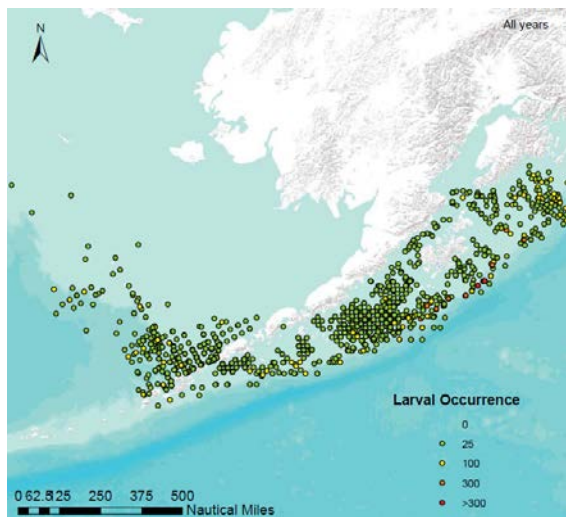


4. Migration: proposed studies

- *Larval migration and connectivity*

Objective: Understand the mechanisms of larval connectivity between the GOA and the BS.

- Collect data from the NMFS ichthyoplankton survey and map larval distribution over time and space.
- Collect larval samples from the survey to conduct genetic analyses.



Collaboration with Janet Duffy-Anderson, Esther Goldstein, William Stockhausen (NOAA-AFSC)

Deliverables:

- Improved knowledge on larval distribution, migration and genetic structure within the population



5. Genetics and genomics: proposed studies

- GENETIC STRUCTURE OF THE POPULATION
- GENOMIC TOOLS (e.g. GENOME)

What is needed?

- Improved knowledge on the genetic composition of the population
- Establish genomic resources for the species
- Genome-wide association studies to evaluate genetic effects of fishery-dependent and fishery-independent influences on growth, reproduction, nutrition, etc.

New proposed studies:

- *Population genetic studies*
- *Sequencing of the Pacific halibut genome*

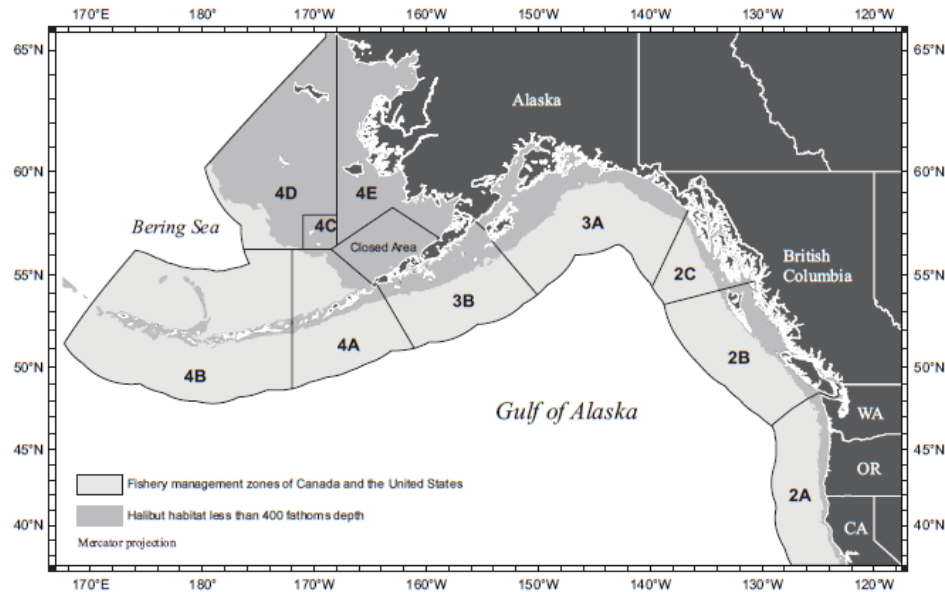


5. Genetics and genomics: proposed studies

- *Population genetic studies*

Objective: Genetic characterization of Pacific halibut throughout its distribution range

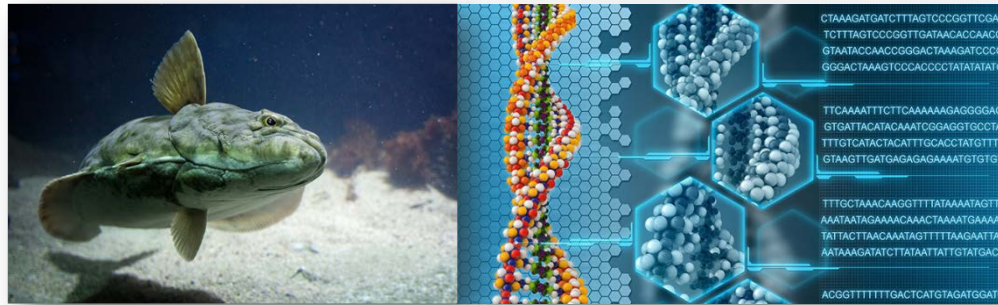
- Characterization of population structure by RAD sequencing and SNP analysis.
- Identification of genetic signatures of geographical population groups



5. Genetics and genomics: proposed studies

- *Pacific halibut genome*

Objective: Obtain a first draft sequence of the Pacific halibut genome



- Identify genomic regions and genes responsible for temporal and spatial adaptive characteristics.
- Genome-wide association studies to try to understand the genetic basis of growth, reproductive performance, migratory behaviour and performance, etc.
- Provide genomic resolution to genetic markers (from RAD tag seq or RNAseq).
Link genotype and phenotype.



Temporal chart of activities

2016	2017	2018	2019	2020	2021
	Annual reproductive cycle				
	Sex determination mechanisms				
Population genetic structure					
	Genome sequencing				
	Genome-wide association studies				
	Epigenome characterization				
Growth transcriptome					
Growth-related patterns					
Regulation of growth by environmental factors					
	Handling practices, injury levels and physiological condition				
	Physiological condition and survival				
Comprehensive studies on migration					
Larval migration and connectivity					
Tagging sublegal halibut					
Reproductive monitoring of PAT-tagged adults					



